The triazine herbicide incidence and the influence of the drying processes on theirs content from maize samples

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Abstract
Atrazine is a pre- or post-emergence herbicide use for the selective control of some annual grasses and most broadleaf weeds in maize, sweet-corn, sorghum, sugarcane, asparagus, vines, top-fruit, citrus, forestry/grassland, etc. The maize metabolizes the atrazine into hydroxyl-atrazine and conjugated amino acids. Atrazine has one of the largest control spectrums; the action period is in the range of 4-6 months. This paper aimed to study the atrazine content detected as hydroxyl-atrazine in maize samples harvested in different ripening phases in the western area of Romania. The influence of the physical drying processes applied on cereals harvest upon the atrazine content was also had in view.

Keywords: maize, atrazine, hydroxyl-atrazine, ripening phases, drying processes

Introduction
In our country, over 500 types of pesticides are applying. The first point of view is about the destruction of the natural enemies of the pests, selection of new types resistant to the pests, modifying of the reproduction process of some species. The second point of view is about the noxiousness of the pesticides for other species excepting the pests: people animals (fish, birds etc.), and plants. For people the risk of acute or chronic intoxication could be of professional, accidental or voluntary nature, as well as by contamination of natural environment – soil, water sources, food, air – having large future implications (Alexa, 2003; Avramides and Gkatsos, 2007; Elkins et. al., 1998; Marcacci, 2004).

Triazine herbicides are derivatives of 4,6-bis-alcilamine 2-chloro-sim-triazine, -2-methoxy-sim-triazine and -2-methyl-mercaptopo-sim-triazine. The main compounds from this class of substances are chlortiazine with the general formula:

\[
\text{NClN} \quad \text{N} \quad \text{N} \quad \text{N} \quad \text{NH}_2 \quad \text{R} \quad \text{NHR}_2
\]

- atrazine (2-Chloro-4-(ethylamino)-6-(isopropylamino)-1,3,5-triazine)
- simazine (2-Chloro-4,6-bis (ethylamino)-1,3,5-triazine)
- propazine (2-Chloro-4,6-bis (isopropylamino)-1,3,5-triazine)

Atrazine represents the most extended compound. It is a selective systemic herbicide, absorbed by roots and leaves, translocated acropetally in the xylem and it accumulates in the apical meristems. It is physiological selective for maize, sorghum species that rapidly
decompounds. This herbicide has one of the largest control spectrums, both the monocotyledonous weeds and the annual dicotyledonous (AOAC, 2006). The action period is in the range of 4-6 months, being possible for it to be applied in autumn, but usually it is applied in spring, replant incorporated, reemergence and post emergence, because it acts on the weeds that are in the stage of germination (Alexa, 2003; Ostafe, 2006).

The action mechanism in weeds fighting is based on the Hill reaction inhibition from the water photolysis process, blocking the photosynthesis process and, finally leading to the plants death. The following crop plants are extremely sensitive to atrazine: rape, mustard, beet, sun-flower, vegetables, tobacco, soybeans, beans, lentil, onion, wheat, barley, oat (Ballantine and Simoneaux, 1991; Raveton, 1996).

The maize metabolizes the atrazine into hydroxyl-atrazine and conjugated amino acids (Ostafe, 2006). Chemical transformation of atrazine into hydroxyl-atrazine has been well studied in maize plant. Hydroxyl metabolites of atrazine have been found in plants grown in soil treated with it, but atrazine itself has not been found in crops (Cantor et. al., 1992). When sprayed on maize, it is quickly transformed by the plant into its hydroxyl metabolites. The replacement of the chlorine atom by a hydroxyl group results in non phytotoxic metabolites and explaining mainly maize tolerance to atrazine (Marcacci 2004).

The atrazine degrades itself quickly in the plants, too: upon a 5kg/ha atrazine enforced on the vegetation treatment, after 10 days, it was found in the maize plants lower than 1,1 ppm. After 3 weeks from this treatment, only amounts lower than 0,1 ppm can be found in plants (Bennett et. al., 2000). In the USA, the residues recorded were lower than 0,06 ppm in carrots, 0,08 ppm in maize and garlic, 0,1 ppm in the soybeans and parsnip roots, 0,34 ppm in sorghum, 0,36 ppm in parsley. For example, the residues amounts were assigned as follows: 0,05-0,24 ppm in the root, 0,05-0,12 ppm in the stem and 0,05-0,8 ppm in the leaves (Alexa, 2003; Ghinea and Vladutiu, 1987; Pussemier et. al., 1997).

**Material and methods**

Maize samples have been analyzed from two counties in the western part of Romania (Arad and Timis) from harvest which have been treated with 5 kg/ha of atrazine. The maize have been harvested in different maturity stages and have been determinate atrazine residues according with *Standard Method-10976 -1990*.

**Atrazine extraction.** 100g sample (corn flour or grounded corn grains) taken according to standards or internal technique norms, was submitted to water and ethylic alcohol extraction for 24 hours and after that, filtered. The obtained filtrate was then submitted to vaporization about 30 minutes at the maximum temperature of 60°C until a volume of 20 mL approximately.

**The atrazine purification.** Carrying on, the resulted solution upon vaporization was submitted to precipitation, proteins separation and fats and waxes action solvent by adding 5cm³ sodium carbonate solution and 50cm³ ethylic ether several times, every time recovering upon decantation (30 minutes leisure) the superior ether fraction. The soaps drops traces remained in the ether solution, were submitted to filtration through the column in which a 5cm Celite 545 layer. The ether extract obtained upon passing through the column with Celite, was submitted to vaporization at 30°C.

**Atrazine hydrolysis.** The obtained residue according to paragraph 2, was quantitatively passed with 10–15cm³ into a separation funnel. Carrying on, the sample was treated with 1cm³ sulphuric acids, homogenized and let to rest for 30 minutes, the operation being repeated three times. After that, 9 ml water have been added in the sample and homogenized. The atrazine hydrolyses passed into aqueous phase.
**Atrazine residues determination by spectrophotometer method.** Atrazine residues determination from the maize samples was performed by spectrophotometer method. After extraction and purification, the atrazine hydrolyses with sulphuric acid, according to the reaction (Figure 1):

![Atrazine and hydroxyl-atrazine](image)

**Figure 1. Atrazine hydrolysis**

The absorbance of hydroxyl-atrazine was detected at 240 nm, 225 nm, 255 nm. The limit minim of detection is 5μg atrazine at 100 g samples (0,05 ppm).

**Colorimetric test.** We measured extinction (absorbance) at the wavelength of 240 nm accountable to a water case. On the same sample it measures the extinction on the wavelength of 225 and 255 nm. The calibration curve was determined between 0,05-0,5 ppm and the linear dynamic range was checked, the average correlation coefficient was 0,945. The mean recovery values ranged from spiked samples at 0,1 ppm was between 85-90%.

**Results and discussions**

The experimental results regarding the content of atrazine as hydroxyl-atrazine (HA) in maize samples as well as the analyzed sample number are presented in Table 1. Samples were taken from counties Arad and Timis belonging to the west part of Romania.

**Table 1 The content of the hydroxy-atrazine in maize samples**

<table>
<thead>
<tr>
<th>County</th>
<th>No. of analyzed samples</th>
<th>Corn maturity stage</th>
<th>Number of positive samples &gt;MAL</th>
<th>Number of undetectable samples &lt;LMD</th>
<th>MAL (ppm)</th>
<th>LMD (ppm)</th>
<th>Number of detectable samples LMD-MAL</th>
</tr>
</thead>
<tbody>
<tr>
<td>AR</td>
<td>10</td>
<td>Crude</td>
<td>-</td>
<td>8</td>
<td>0.1</td>
<td>0.05</td>
<td>2</td>
</tr>
<tr>
<td></td>
<td>10</td>
<td>Yellow</td>
<td>-</td>
<td>9</td>
<td>0.1</td>
<td>0.05</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>10</td>
<td>Complete</td>
<td>-</td>
<td>10</td>
<td>0.1</td>
<td>0.05</td>
<td>-</td>
</tr>
<tr>
<td>TM</td>
<td>10</td>
<td>Crude</td>
<td>-</td>
<td>10</td>
<td>0.1</td>
<td>0.05</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>10</td>
<td>Yellow</td>
<td>-</td>
<td>10</td>
<td>0.1</td>
<td>0.05</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>10</td>
<td>Complete</td>
<td>-</td>
<td>10</td>
<td>0.1</td>
<td>0.05</td>
<td>-</td>
</tr>
</tbody>
</table>

*MAL – maximum admitted limit*  
*LMD – minim detection limit*  
*AR – Arad County*  
*TM – Timisoura County*
Table 2 Moisture variation of corn grains in different maturity stages

<table>
<thead>
<tr>
<th>Ripening phases</th>
<th>Moisture, (%)</th>
<th>Dried substance (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Crude (early)</td>
<td>84.6</td>
<td>15.4</td>
</tr>
<tr>
<td>Crude (middle)</td>
<td>66.9</td>
<td>33.1</td>
</tr>
<tr>
<td>Crude (late)</td>
<td>48.7</td>
<td>51.3</td>
</tr>
<tr>
<td>Yellow (early)</td>
<td>36.7</td>
<td>63.3</td>
</tr>
<tr>
<td>Yellow (middle)</td>
<td>27.9</td>
<td>72.1</td>
</tr>
<tr>
<td>Complete</td>
<td>17.0</td>
<td>83</td>
</tr>
</tbody>
</table>

Table 3 The classification of maize sample according the moisture content (%) (Alexa, 2003)

<table>
<thead>
<tr>
<th>Crop</th>
<th>Dry (%)</th>
<th>Semidry (%)</th>
<th>Humid (%)</th>
<th>Very humid (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Maize</td>
<td>12 - 14</td>
<td>14.1 - 17</td>
<td>17.1 - 20</td>
<td>above 20</td>
</tr>
</tbody>
</table>

The obtained experimental results regarding the determinate atrazine residues with hydroxyl-atrazine content did not record positive values at no of the analyzed samples, but the obtained values had situated in proportion of 20% between 0.05-0.1 ppm, in the case of the samples early harvest in milk phase coming from AR county and in 10% in the samples harvest in yellow early phases.

In Timis county was not registered positive values regarding the atrazine content in neither of the analyzed samples, no matter of the corn maturity stage.

For the assurance of adequate preservation of cereals in bunkers, the moisture which exceed normal limit (14-15%) is extracted through the dried procedure. Through drying we follow the remove the water physical banded, process which is realized through the phenomenon of internal and external water diffusion from grain interior to the exterior.

The corn drying it realized through convection, through the transmission of the heat in grains mass by the warm air or gases combustion in mixture with atmospheric air using not very high drying temperatures (110 – 130°C). The proportion between the burning gases and atmospheric air is 1:20, in winter and 1:30.

Experimental, grounded maize samples harvested in the full maturity stage spiked with atrazine in 1 ppm amount, was dried according to drying temperatures practiced in the cereals storage and the influence of the drying on the atrazine content from the contaminated samples was aimed.

The maize drying was carried out at the 100-130°C temperatures. At drying temperatures that surpass these limits, the maize grains fissure. The initial moisture content of samples was between 17-19% and the drying process was guided between 100-130°C until at 14% final moisture.

The amount of atrazine was pursued on the way to the drying process (Figure 2).
Figure 2 The influence of drying process on hydroxyl-atrazine content and moisture

Conclusions
Atrazine content determinate as hydroxyl-atrazine, is framing in the maximum limit allowed for all the analyzed samples, unaffected by the corn maturity stage, do not exist any contamination with triazine compounds;
The samples taken from Timis county was in the range of undetectable values, 3 samples coming from AR county, early harvest in milk or yellow phase, are situated in the range of detectable values, but without exceeding maximum admitted limit, with values in the range of 0,05-0,1 ppm.
The maize crop drying at 100-130°C temperatures does not lead to significant reduction of the hydroxyl-atrazine content from the maize sample.

References
Bennett E.R., Moore M.T., Cooper C.M., (2000), Method for the simultaneous extraction and analysis of two current use pesticides, atrazine and lambda-cyhalothrin, in sediment and aquatic plants, Bull Environ Contam Toxicol, 64, 825-833.


